

**PM1-AHAS1 nucleotide sequence(SEQ ID NO: 1) and
translated amino acid sequence (SEQ ID NO: 101)**

1 TCATCATCTCTCTCTCTCAAAACCATGGCGGCGGCAACATCGTCTTCTCCGATCTCCTTAACCGCTAAACCTTCTTCCAAATCCCCTCTACCCATTTC
M A A A T S S S P I S L T A K P S S K S P L P I S

101 GATTCTCCCTTCCCTTCTCCTTAACCCACAGAAAGACTCCTCCCGTCTCCACCGTCTCTCGCCATCTCCGCCGTTCTCAACTCACCCGTCAATGTTCGC
R F S L P F S L T P Q K D S S R L H R P L A I S A V L N S P V N V A

201 ACCTCCTTCCCCTGAAAAAACCGACAAGAACAAGACTTTCGTCTCCCGCTACGCTCCCGACGAGCCCCGAAGGGTGCTGATATCCTCGTCGAAGCCCTC
P P S P E K T D K N K T F V S R Y A P D E P R K G A D I L V E A L

301 GAGCGTCAAGGCGTCGAAACCGTCTTTGCTTATCCCGGAGGTGCTTCCATGGAGATCCACCAAGCCTTGACTCGCTCCTCCACCATCCGTAAACGTCCTTC
E R Q G V E T V F A Y P G G A S M E I H Q A L T R S S T I R N V L

401 CCCGTCACGAACAAGGAGGAGTCTTCGCCGCCGAGGGTTACGCTCGTTCTCCGGCAAACCGGAATCTGCATAGCCACTTCGGGTCGCCGAGCTACCAA
P R H E Q G G V F A A E G Y A R S S G K P G I C I A T S G P G A T N

501 CCTCGTCAGCGGTTAGCAGACGCGATGCTTGACAGTGTTCTCTTGTCGCCATTACAGGACAGGTCCCTCGCCGGATGATCGGTACTGACGCCTTCCAA
L V S G L A D A M L D S V P L V A I T G Q V P R R M I G T D A F Q

601 GAGACACCAATCGTTGAGTAACGAGGTCTATTACGAAACATAACTATCTGGTGATGGATGTTGATGACATACCTAGGATCGTTCAAGAAGCATTCTTTC
E T P I V E V T R S I T K H N Y L V M D V D D I P R I V Q E A F F

701 TAGCTACTTCCGGTAGACCCGGACCGGTTTTGGTTGATGTTCTTAAGGATATTAGCAGCAGCTTGCATTCTTAAGTGGATCAACCTATGCGCTTGCC
L A T S G R P G P V L V D V P K D I Q Q Q L A I P N W D Q P M R L P

801 TGGCTACATGTCTAGGTTGCCTCAGCCWCCGGAAGTTTCTCAGTTAGGTGATCGTTAGGTTGATCTCGGAGTCTAAGAGGCCTGTTTTGTACGTTGGT
G Y M S R L P Q X P E V S Q L G Q I V R L I S E S K R P V L Y V G

901 GGTGGAAGCTTGAAGTCAAGTGAAGAAGTGGGAGATTGTGCGAGCTTACTGGGATCCCTGTTGCGAGTACGTTGATGGGGCTTGGCTCTTATCCTTGTA
G G S L N S S E E L G R F V E L T G I P V A S T L M G L G S Y P C

1001 ACGATGAGTTGTCCCTGCAGATGCTTGGCATGCACGGGACTGTGTATGCTAACTACGCTGTGGAGCATAGTGATTGTTGCTGGCGTTTGGTGTAGGTT
N D E L S L Q M L G M H G T V Y A N Y A V E H S D L L L A F G V R F

1101 TGATGACCGTGTACGGGAAAGCTCGAGGCTTTCTCGTAGCAGGGCTAAAATGTGCACATAGACATTGATTCTGCTGAGATTGGGAAGAATAAGACACCT
D D R V T G K L E A F A S R A K I V H I D I D S A E I G K N K T P

1201 CACGTGTCTGTGTGTGGTGATGTAAAGCTGGCTTTGCAAGGGATGAACAAGGTTCTTGAGAACCGGGCGGAGGAGCTCAAGCTTGATTTCCGTGTTTGGGA
H V S V C G D V K L A L Q G M N K V L E N R A E E L K L D F G V W

1301 GGAGTGAGTTGAGCGAGCAGAAACAGAAGTTCCCTTTGAGCTTCAAAACGTTTGGAGAAGCCATTCTCCGAGTACGCGATTGAGATCCTCGACGAGCT
R S E L S E Q K Q K F P L S F K T F G G E A I P P Q Y A I Q I L D E L


1401 AACCGAAGGGAAGGCAATTATCAGTACTGGTGTGGACAGCGTCAGATGTGGGCGGCGCAGTTTACAAGTACAGGAAGCCGAGACAGTGGCTGTCTGCA
T E G K A I I S T G V G Q R Q M W A A Q F Y K Y R K P R Q W L S S

1501 TCAGGCCTCGGAGCTATGGGTTTGGACTTCTCTGCTGCGATTGGAGCGTCTGTGGCGAACCTGATGCGATTGTTGTGGATATTGACGGTGATGGAAGCT
S G L G A M G F G L P A A I G A S V A N P D A I V V D I D G D G S

1601 TCATAATGAACGTTCAAGAGCTGGCCACAATCCGTGTAGAGAATCTTCTGTGAAGATACTCTGTAAACAACAGCATCTTGGGATGGTCATGCAATG
F I M N V Q E L A T I R V E N L P V K I L L L N N Q H L G M V M Q W

1701 GGAAGATCGGTTCTACAAAGCTAACAGAGCTCACACTTATCTCGGGGACCCGGCAAGGGAGAACGAGATCTTCCCTAACATGCTGCAGTTTGCAGGAGCT
E D R F Y K A N R A H T Y L G D P A R E N E I F P N M L Q F A G A

1801 TCGGGGATTCCAGCTGCGAGAGTGACGAAGAAAGAAGAACTCCGAGAAGCTATTACAGACAATGCTGGATACACCAGGACCATACTGTTGGATGTGATAT
C G I P A A R V T K K E E L R E A I Q T M L D T P G P Y L L D V I

1901 GTCCGCACCAAGAACATGTGTTACCGATGATCCCAATGGTGGCACTTTCAAAGATGTAATAACAGAAGGGGATGGTCGCACTAAGTACTGAGAGATTMA
C P H Q E H V L P M I P  G G T F K D V I T E G D G R T K Y

2001 GCTGGTGATCGATCATATGGTAAAAGACTTAGTTTCAGTTTCCAGTTTCTTTTGTGTGGTAATTTGGGTTTGTGAGTTGTTGT

Figure 1A

**PM2-AHAS3 nucleotide sequence (SEQ ID NO:2) and
translated amino acid sequence (SEQ ID NO: 102)**

1 TTCATCATMTCTCTCTCATTTCTCTCTCTCTCATCTAACCATGGCGGCGGCAACATCGTCTTCTCCGATCTCCTTAACCGCTAAACCTTCTTCCAAAT
M A A A T S S S P I S L T A K P S S K

101 CCCCTCTACCCATTTCCAGATTCTCCCTTCCCTTCTCCTTAACCCACAGAAACCTCCTCCCGTCTCCACCGTCCACTCGCCATCTCCGCCGTTCTCAA
S P L P I S R F S L P F S L T P Q K P S S R L H R P L A I S A V L N

201 CTCACCCGTCAATGTCGCACCTGAAAAAACCGACAAGATCAAGACTTTTCATCTCCCGCTACGCTCCCGACGAGCCCCGAAGGGTGCTGATATCCTCGTG
S P V N V A P E K T D K I K T F I S R Y A P D E P R K G A D I L V

301 GAAGCCCTCGAGCGTCAAGGCGTCGAAACCGTCTTCGCTTATCCCGAGGTGCCTCCATGGAGATCCACCAAGCCTTGACTCGCTCCTCCACCATCCGTA
E A L E R Q G V E T V F A Y P G G A S M E I H Q A L T R S S T I R

401 ACGTCTCCCCCGTCAAGAACAGGAGAGTCTTCGCCCGGAGGGTTACGCTCGTTCCTCCGGCAAACCGGAATCTGCATAGCCACTTCGGGTCCCGG
N V L P R H E Q G G V F A A E G Y A R S S G K P G I C I A T S G P G

501 AGCTACCAACCTCGTCAGCGGGTTAGCCGACGCGATGCTTGACAGTGTTCCTCTCGTCGCCATCACAGGACAGGTCCCTCGCCGGATGATCGGTACTGAC
A T N L V S G L A D A M L D S V P L V A I T G Q V P R R M I G T D

601 GCGTTCCAAGAGACGCCAATCGTTGAGGTAACGAGGTCTATTACGAAACATAACTATCTGGTGATGGATGTTGATGACATACCTAGGATCGTTCAAGAAG
A F Q E T P I V E V T R S I T K H N Y L V M D V D D I P R I V Q E

701 CATTCTTTCTAGTACTTCCGGTAGACCCGGACCGGTTTGGTTGATGTTCTCTAAGGATATTAGCAGCAGCTTGCGATTCTTAAGTGGATCAACCTAT
A F F L A T S G R P G P V L V D V P K D I Q Q Q L A I P N W D Q P M

801 GCGCTTGCCTGGCTACATGTCTAGGCTGCCTCAGCCACCGGAAGTTTCTCAGTTAGGCCAGATCGTTAGGTTGATCTCGGAGTCTAAGAGCCCTGTTTTG
R L P G Y M S R L P Q P P E V S Q L G Q I V R L I S E S K R P V L

901 TACGTTGGTGGTGGAAGCTTGAAGTTCGAGTGAAGAACTGGGGAGATTGTGCGAGCTTACTGGGATCCCTGTTGCGAGTACGCTGATGGGGCTTGGCTCTT
Y V G G G S L N S S E E L G R F V E L T G I P V A S T L M G L G S

1001 ATCCTTGTAACGATGAGTTGTCCCTGCAGATGCTTGGCATGCACGGGACTGTGTATGCTAACTACGCTGTGGAGCATAGTGATTGTTGCTGGCGTTTGG
Y P C N D E L S L Q M L G M H G T V Y A N Y A V E H S D L L L A F G

1101 TGTTAGGTTTGTATGACCGTGTACGGGAAAGCTCGAGGCGTTTTCGAGCAGGGCTAAGATTGTGCACATAGACATTGATTCTGCTGAGATTGGGAAGAAT
V R F D D R V T G K L E A F A S R A K I V H I D I D S A E I G K N

1201 AAGACACCTCAGTGTCTGTGTGTGGTGTGTAAAGCTGGCTTTGCAAGGGATGAACAAGGTTCTTGAGAACCGGGCGGAGGAGCTCAAGCTTGATTTTCG
K T P H V S V C G D V K L A L Q G M N K V L E N R A E E L K L D F

1301 GTGTTTGGAGGAGTGAGTTGAGCGAGCAGAAACAGAAGTTCCTGTTGAGCTTCAAAACGTTTGGAGAAGCCATTCTCCGCGAGTACGCGATTACAGGTCCT
G V W R S E L S E Q K Q K F P L S F K T F G E A I P P Q Y A I Q V L

1401 AGACGAGCTAACCCAGGGAAGGCAATTATCAGNACTGGTGTGGACAGCATCAGATGTGGGCGGCGAGTTTACAAGTACAGGAAGCCGAGGCGAGTGG
D E L T Q G K A I I X T G V G Q H Q M W A A Q F Y K Y R K P R Q W

1501 CTGTCGTCCTCAGGACTCGGAGCTATGGGTTTTCGACTTCCTGCTGCGATTGGAGCGTCTGTGGCGAACCCTGATGCGATTGTTGTGGACATTGACGGTG
L S S S G L G A M G F G L P A A I G A S V A N P D A I V V D I D G

1601 ATGGAAGCTTCATAATGAACGTTCAAGAGCTGGCCACAATCCGTGTAGAGAATCTTCTGTGAAGATACTCTTGTAAACAACCAGCATCTTGGGATGGT
D G S F I M N V Q E L A T I R V E N L P V K I L L L N N Q H L G M V

1701 CATGCAATTGGAAGATCGGTTCTACAAAGCTAACAGAGCTCACACTTATCTCGGGGACCCGGCAAGGGAGAACGAGATCTTCCCTAACATGCTGCAGTTT
M Q L E D R F Y K A N R A H T Y L G D P A R E N E I F P N M L Q F

1801 GCAGGAGCTTGCGGGATTCCAGCTGCGAGAGTGACGAAGAAAGAAGAACTCCGAGAAGCTATTACAGACAATGCTGGATACACCTGGACCGTACCTGTTGG
A G A C G I P A A R V T K K E E L R E A I Q T M L D T P G P Y L L

1901 ATGCCATCTGTCCGACCAAGAACATGTGTTACCGATGATCCCAAGTGGTGGCACTTTCAAAGATGTAATAACCGAAGGGGATGGTTCGCACTAAGTACTG
D A I C P H Q E H V L P M I P S G G T F K D V I T E G D G R T K Y

2001 AGAGATGAAGCTGGTGATCCATCGTATGGTAAAAGACTTAGTTTCAGTTTTCAGTTTCTTTGTGTGGTAATTTGGGTTTGTGAGTTGTTGTTGTGCTTT

2101 TGGTTTGTTCCKNAC

Figure 1B

Figure 1C

**T-AHAS3 (SEQ ID NO: 4) nucleotide sequence and
translated amino acid sequence (SEQ ID NO: 104)**

1 TTMMACATCTCTCTCATTNCACTCTCTCCCTCATCTAACCATGGCGGGCGGCAACATCGCCTTCTCCGATCTCCTTAACCGCTAAACCTTCTTCCAAAT
M A A A T S P S P I S L T A K P S S K

101 CCCCTCTACCCATTTCCAGATTCTCCCTTCCCTTCTCCTTAACCCACAGAAACCTCTCCCGTCTCCACCGTCCACTCGCCATCTCCGCCGTTCTCAA
S P L P I S R F S L P F S L T P Q K P S S R L H R P L A I S A V L N

201 CTCACCCGTCAATGTGCGACCTGAAAAACCGACAAGATCAAGACTTTTCATCTCCCGCTACGCTCCCGACGAGCCCCGCAAGGGTGCTGATATCCTCGTG
S P V N V A P E K T D K I K T F I S R Y A P D E P R K G A D I L V

301 GAAGCCCTCGAGCGTCAAGGCGTCAAGAACCGTCTTCGCTTATCCCGGAGGTGCCTCCATGGAGATCCACCAAGCCTTGACTCGCTCCTCCACCATCCGTA
E A L E R Q G V E T V F A Y P G G A S M E I H Q A L T R S S T I R

401 ACGTCTCCCCCGTCACGAACAAGGAGGAGTCTTCGCGCGCGAGGGTTACGCTCGTTCCTCCGGCAAACCGGAATCTGCATAGCCACTTCGGGTCCCGG
N V L P R H E Q G G V F A A E G Y A R S S G K P G I C I A T S G P G

501 AGCTACCAACCTCGTCAGCGGGTTAGCCGACGCGATGCTTGACAGTGTTCCTCTCGTCGCCATCACAGGACAGGTCCCTCGCCGGATGATCGGTACTGAC
A T N L V S G L A D A M L D S V P L V A I T G Q V P R R M I G T D

601 GCGTTCCAAGAGACGCCAATCGTTGAGGTAAACGAGGTCTATTACGAAACATAACTATCTGGTGATGGATGTTGATGACATACCTAGGATCGTTCAAGAAG
A F Q E T P I V E V T R S I T K H N Y L V M D V D D I P R I V Q E

701 CATTCTTTCTAGCTACTTCCGGTAGACCCGGACCGGTTTTGGTTGATGTTCTTAAGGATATTCAGCAGCAGCTTGCGATTCTTAAGTGGATCAACCTAT
A F F L A T S G R P G P V L V D V P K D I Q Q Q L A I P N W D Q P M

801 GCGCTTGCTGGTACATGTCTAGGCTGCCTCAGCCACCGGAAGTTTCTCAGTTAGGCCAGATCGTTAGGTTGATCTCGGAGTCTAAGAGGCCTGTTTTG
R L P G Y M S R L P Q P P E V S Q L G Q I V R L I S E S K R P V L

901 TACGTTGGTGGTGAAGCTTGAACCTCGAGTGAGGAACTGGGGAGATTGTGCGAGCTTACTGGGATCCCTGTTGCGAGTACGTTGATGGGGCTTGGCTCTT
Y V G G G S L N S S E E L G R F V E L T G I P V A S T L M G L G S

1001 ATCCTTGTAACGATGAGTTGTCCCTGCAGATGCTTGGCATGCACGGGACTGTGTATGCTAACTACGCTGTGGAGCATAGTGATTGTTGCTGGCGTTTGG
Y P C N D E L S L Q M L G M H G T V Y A N Y A V E H S D L L L A F G

1101 TGTTAGGTTTGATGACCGTGTACGCGGAAAGCTCGAGGCGTTTGCAGACAGGGCTAAGATTGTGCACATAGACATTGATCTGCTGAGATTGGGAAGAAT
V R F D D R V T G K L E A F A S R A K I V H I D I D S A E I G K N

1201 AAGACACCTCAGTGTCTGTGTGGTGTGTAAGCTGGCTTTGCAAGGGATGAACAAGGTTCTTGAGAACCGGGCGGAGGAGCTCAAGCTTGATTTGCG
K T P H V S V C G D V K L A L Q G M N K V L E N R A E E L K L D F

1301 GTGTTTGGAGGAGTGAGTTGAGCGAGCAGAAACAGAAGTTCCCGTTGAGCTTCAAACGTTTGGAGAAGCCATTCTCCGAGTACGCGATTACAGTCTCT
G V W R S E L S E Q K Q K F P L S F K T F G E A I P P Q Y A I Q V L

1401 AGACGAGCTAACCCAAAGGGAAGCAATTATCAGTACTGGTGTGGACAGCATCAGATGTGGCGCGCAGTTTACAAGTACAGGAAGCCGAGGCAGTGG
D E L T Q G K A I I S T G V G Q H Q M W A A Q F Y K Y R K P R Q W

1501 CTGTCGTCCTCAGGACTCGGAGCTATGGGTTTTCGGACTTCTGCTGCGATTGGAGCGTCTGTGGCGAACCCTGATGCGATTGTTGTGGACATTGACGGTG
L S S S G L G A M G F G L P A A I G A S V A N P D A I V V D I D G

1601 ATGGAAGCTTCATAATGAACGTTCAAGAGCTGGCCACAATCCGTGTAGAGAATCTTCCTGTGAAGATACTCTTGTTAAACAACCAGCATCTTGGGATGGT
D G S F I M N V Q E L A T I R V E N L P V K I L L L N N Q H L G M V

1701 CATGCAATGGGAAGATCGGTTTACAAAGCTAACAGAGCTCACACTTATCTCGGGGACCCGCAAGGGAGAACGAGATCTTCCCTAACATGCTGCACTTT
M Q W E D R F Y K A N R A H T Y L G D P A R E N E I F P N M L Q F

1801 GCAGGAGCTTTCGGGATTCAGCTGCGAGAGTGACGAAGAAAGAACTCCGAGAAGCTATTACAGACAATGCTGGATACACCTGGACCGTACCTGTTGG
A G A C G I P A A R V T K K E E L R E A I Q T M L D T P G P Y L L

1901 ATGTCTCTGTCCGACCAAGAACATGTGTTACCGATGATCCCAAGTGGTGGCACTTTTCAAGATGTAATAACCGAAGGGGATGGTCCGACTAAGTACTG
D V I C P H Q E H V L P M I P S G G T F E D V I T E G D G R T K Y

2001 AGAGATGAAGCTGGTGTATCCATCATATGGTAAAAGACTTAGTTTCAGTTTACAGTTTCTTTGTGTGGTAATTTGGGTTTGTGAGTTGTTCTGCTTT

2101 TGGTTTGTTCCKWAC

Figure 1D

List of oligonucleotides suitable for detection of the PM1 and PM2 mutations. All oligonucleotides are in 5'-3' orientation.

SEQ ID NO.	AHAS1-PM1 forward	SEQ ID NO.	AHAS1-PM1 reverse	SEQ ID NO.	AHAS3-PM2 forward	SEQ ID NO.	AHAS3-PM2 reverse
5	TTATCTCGGGACCGGCAA	24	CATCTTTGAAGTGCACCA	47	CTCAGGACTCGGAGCTATGG	66	CTTTGTAGAACCGATCTTCC
6	GACCCGGCAAGGGAGAACGA	25	TCGTATTATCATCTTTGAA	48	GGAGCTATGGTTTCGGACT	67	GCTCTGTTAGCTTTGTAGAA
7	GGGAGAACGAGATCTTCCT	26	ACCATCCCTTCTGTATTAA	49	GTTTCGGACTCTCTGCTCGG	68	ATAAGTGTGAGCTCTGTTAG
8	GATCTTCCCTAACATGCTGC	27	ACTTAGTCGACCACTCCCT	50	TCCTGTGCGATTGGAGCGT	69	GGTCCCCGAGATAAGTGTA
9	AACATGCTGCAGTTTGCAGG	28	ATCTCTCAGTACTTAGTGCG	51	ATTGGAGCGTCTGTGGCGAA	70	TCCCTTGC CGGTCCCGAG
10	AGTTTGCAGGAGCTTGCAGG	29	CACGAGCTTCATCTCTCAGT	52	CTGTGGCGAACCCCTGATCGG	71	GATCTGTTCTCCCTTGCCG
11	AGCTTGGGGATTCAGCTG	30	TATGATCGATCACCACTTC	53	CCCTGATGCGATTGTTGG	72	TGTTAGGGAAGATCTCGTTC
12	ATTCCAGCTGGGAGAGTGAC	31	TCCTTTACCATATGATCGAT	54	ATTGTTGTGCACATGTGCGG	73	AACGACGATGTTAGGGAA
13	CGAGAGTGACGAAGAAAGAA	32	TGAAACTAAGTCTTTTACCA	55	ACATTGACGGTGATGGAAAGC	74	AGCTCCTGCAAACTGCAGCA
14	GAAGAAAGAACTCCGAG	33	AACGGAACCTGAAACTAAG	56	TGATGGAAGCTTCAATGA	75	GAATCCCGCAAGCTCCTGCA
15	GAATCCGAGAAGCTATTCA	34	ACACAAAAGAACTGGAAC	57	TTCATAATGAACGTTCAAGA	76	CTCGAGCTGGAATCCCGCA
16	AAGCTATTACAGCAATGCTG	35	CCAAATTACCAACACAAAAGA	58	ACGTTCAAGAGCTGGCCACA	77	CTTCGTCACTCTCGCAGCTG
17	GACAATGCTGGATACACCAG	36	ACTGACAAACCCAAATTACC	59	GCTGGCCACAATCCGTGTAG	78	GTTCCTTCTTCTTCGTCACT
18	GATACACGAGACCATACCT	37	TAGTACAACTGACAPAC	60	ATCCGTGTAGAGAACTTCC	79	GCTTCTCGGAGTCTTCTTT
19	GACCATACCTGTTGGATGTG	38	CAACCAAAAGTAGTACAACA	61	AGAACTCTTCTGTGAAGATA	80	TGCTGAAATAGCTTCTCGGA
20	GTTGGATGTATATGTCGCG	39	CGTCTGGGAACAACCAAAAG	62	TGTGAAGATACTCTTGTAA	81	TATCCAGCATTTGTCTGAATA
21	ATATGTCGCGACCAAGAACAA	40	ACAGCGAGTAGCTCTGGGAA	63	CTCTTGTTTAAACAAACCAACA	82	GGTCCAGGTGTATCCAGCAT
22	ACCAAGAACATGTGTTACCG	41	CAAAACAACACAGCGAGTA	64	ACAACAGCATCTTTGGGATG	83	CAACAGGTACGGTCCAGGTG
23	TGTGTTACCGATGATCCCAA	42	AAAAAGGAAACAAACAAACA	65	TCTTGGGATGGTTCATGCAAT	84	AGATGACATCCAAACAGGTAC
43	ATGATCCCAAGTGGTGCACT	44	AGTGCCACCACTTGGGATCAT	85	GTCATGCAATGGGAAGATCGG	86	CCGATCTTCCCATTTGCATGAC
45	ATGATCCCAAAATGGTGCACT	46	AGTGCCACCAATTTGGGATCAT	87	GTCATGCAATTTGGAAGATCGG	88	CCGATCTTCCCAATTTGCATGAC

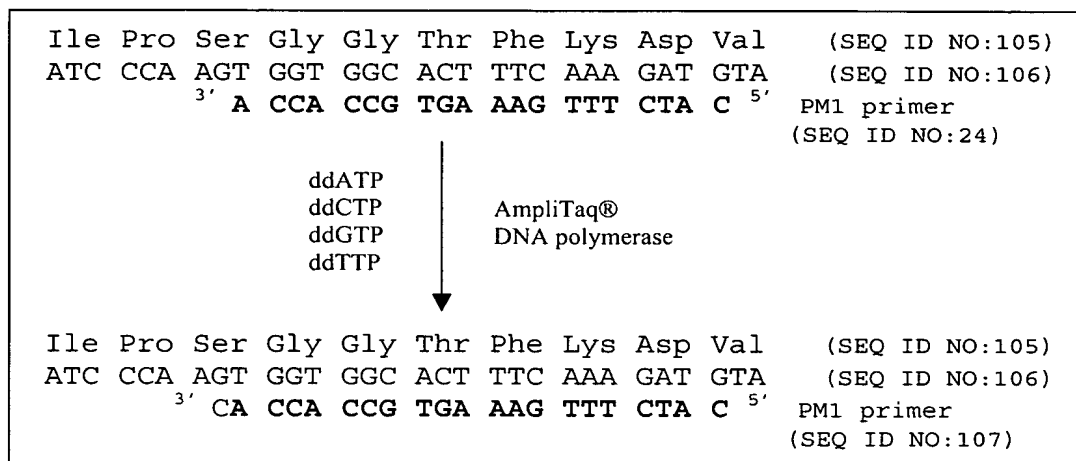
All oligonucleotides noted as being in the forward direction are located on the 5' side of either PM1 or PM2 mutation, in reference to SEQ ID:1 and SEQ ID:2. All oligonucleotides noted as being in the reverse direction are located on the 3' side of either the PM1 or PM2 mutation, in reference to SEQ ID:3 and SEQ ID:4.

Figure 1E

“PM1” Test

AHAS1

‘Topas’



‘PM1’

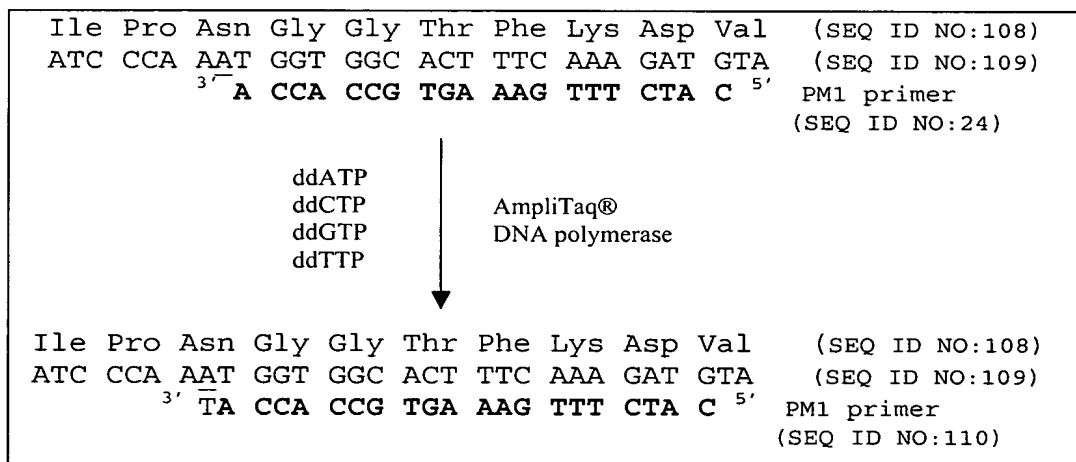
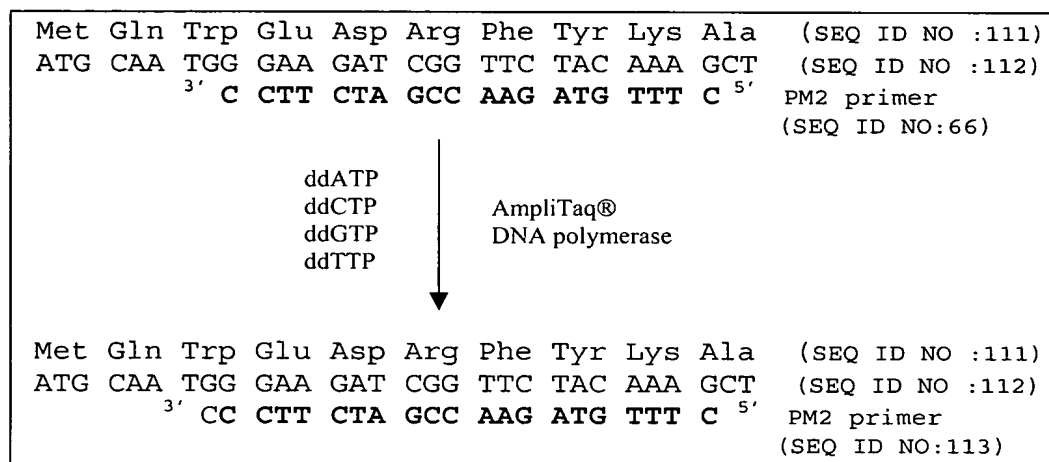


Figure 2

“PM2” Test

AHAS3

‘Topas’



‘PM2’

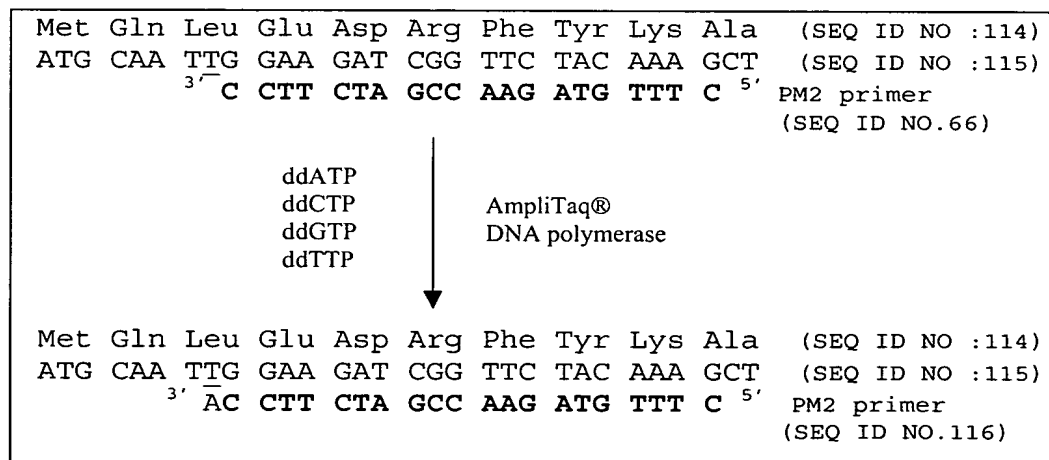


Figure 3

Doubled Haploid Canola Lines		
Plant number	class	GH Rating
1	"PM1/PM2"	0
2	"PM1/PM2"	0
3	"PM1/PM2"	0
4	"PM1/PM2"	0
6	"PM1/PM2"	5
7	"PM1/PM2"	5
37	"PM2"	20
38	"PM2"	20
39	"PM2"	20
40	"PM2"	25
41	"PM2"	25
42	"PM2"	25
68	"PM1"	40
69	"PM1"	40
70	"PM1"	40
71	"PM1"	40
72	"PM1"	40
73	"PM1"	45
103	WT	80
104	WT	80
105	WT	80
106	WT	80
107	WT	85
108	WT	85
-	"PM1"	-
-	"PM2"	-
-	WT	-

Figure 4

Summary of results from “PM1” and “PM2” mutation tests with three replicates using the ABI PRISM® Technology

DNA class	Plant number ²	“PM1” mutation test			“PM2” mutation test		
		Observed ³	Expected ⁴	“PM1” mutation	Observed ³	Expected ⁴	“PM2” mutation
“PM1/PM2” class	1	(C)- <i>T</i>	C- <i>T</i>	yes	(C)- <i>A</i>	C- <i>A</i>	yes
	2	(C)- <i>T</i>	C- <i>T</i>	yes	(C)- <i>A</i>	C- <i>A</i>	yes
	3	(C)- <i>T</i>	C- <i>T</i>	yes	(C)- <i>A</i>	C- <i>A</i>	yes
	4	(C)- <i>T</i>	C- <i>T</i>	yes	(C)- <i>A</i>	C- <i>A</i>	yes
	6	(C)- <i>T</i>	C- <i>T</i>	yes	(C)- <i>A</i>	C- <i>A</i>	yes
	7	(C)- <i>T</i>	C- <i>T</i>	yes	(C)- <i>A</i>	C- <i>A</i>	yes
“PM2” class	37	(C)- <i>T</i>	C	yes	(C)- <i>A</i>	C- <i>A</i>	yes
	38	(C)- <i>T</i>	C	yes	C- <i>A</i>	C- <i>A</i>	yes
	39	(C)- <i>T</i> -(A)	C	yes	(C)- <i>A</i>	C- <i>A</i>	yes
	40	C	C	no	(C)- <i>A</i>	C- <i>A</i>	yes
	41	(C)- <i>T</i>	C	yes	(C)- <i>A</i>	C- <i>A</i>	yes/no
	42	(C)- <i>T</i>	C	yes	(C)- <i>A</i>	C- <i>A</i>	yes
“PM1” class	68	<i>T</i>	C- <i>T</i>	yes	C	C	no
	69	<i>T</i>	C- <i>T</i>	yes	C	C	no
	70	<i>T</i>	C- <i>T</i>	yes	C	C	no
	71	<i>T</i>	C- <i>T</i>	yes	C	C	no
	72	<i>T</i>	C- <i>T</i>	yes	C	C	no
	73	<i>T</i>	C- <i>T</i>	yes	C	C	no
“WT” class	103	C	C	no	C	C	no
	104	C	C	no	C	C	no
	105	C	C	no	C	C	no
	106	C	C	no	C	C	no
	107	C	C	no	C	C	no
	108	C	C	no	C	C	no
‘PM1’ control ¹	-	(C)- <i>T</i>	C- <i>T</i>	yes	C	C	no
‘PM2’ control ¹	-	C	C	no	C- <i>A</i>	C- <i>A</i>	yes
‘Topas’ (WT) control ¹	-	C	C	no	C	C	no

¹PM1’ control and ‘PM2’ control are DNA isolated from “PM1” and “PM2” plants used to develop the tests.

²This number refers to the plant number for DH line identified in Figure 4

³In **Bold** and *italics* are the peaks related to the mutations and in brackets are the peaks that are not always present in all the three replicates.

⁴Expected results assuming that the AHAS amplification reaction using AHAS1/AHAS3 amplification primers (SED ID NO:90 and 91) amplified similar amounts of both *AHAS1* and *AHAS3* sequences and that the PM1 extension primers will anneal also to the *AHAS3* sequence and the PM2 extension primers will anneal also to the *AHAS1* sequence.

Figure 5

Summary of results from “PM1” and “PM2” mutation tests using the Pyrosequencing PSQ™96 Technology

DNA sample	“PM1” mutation test		“PM2” mutation test		SEQ ID NO	“PM2” Mutation
	Pyrosequencing results	SEQ ID NO	Pyrosequencing results	“PM1” Mutation		
‘PM1’	CAAA TGGTGG	98	GGGAAAGATC	Yes	99	No
‘PM2’	CAAGTGGTGG	97	TGGAAAGATC	No	100	Yes
‘Topas’ (WT)	CAAGTGGTGG	97	GGGAAAGATC	No	99	No

FIGURE 6